FINAL REPORT

Microbiological Sampling Report

for

National Oceanic & Atmospheric Administration

Samplings Conducted on the Tenth Floor
of Building SSMC-3
on February 17, 2000

Interagency Agreement #: D8H00CO31200
Task: 9903

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Prepared by

US Public Health Service

Division of Federal Occupational Health

Bethesda Central Office

Executive Summary

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 10129 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 10817. Air samples were also collected from outdoors.

Findings are as follows:

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, contact plate, and dust samples.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Higher fungal levels were detected from samples collected from horizontal surfaces in room 10129 than those in reference room 10817.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in plenum dust of reference room was higher than that of room 10129 (10^5 vs. 10^4 CFU/g of fine dust). *Penicillium* and *Aspergillus* (*Aspergillus niger* included) dominated these samples.

· Fungal levels in furniture dust of these rooms were at 10^3 CFU/g levels.

INTRODUCTION

At the request of the National Oceanic & Atmospheric Administration (NOAA), Federal Occupational Health (FOH) conducted a microbiological sampling in a room 10129 of Building SSMC-3, located at 1315 East-West Highway, Silver Spring, Maryland. Sampling was conducted on February 17, 2000. Air (both Andersenâ and Zefonâ), swab, contact plate, and vacuum dust samples were collected from this room and an indoor reference room 10817. Air samples were also collected from outdoors.

EVALUATION METHODOLOGY

Air Samples

Various types of samples were collected from these rooms on February 17, 2000. Two types of air samples were collected from each room: (1) culturable method using Andersenâ N-6 samplers at a flow rate of 28.3 L/min, and (2) non-culturable method using Zefonâ Air-O-Cell cassettes at a flow rate of 15 L/min. Indoor Andersenâ air samples were collected for 3 minutes and outdoor samples were collected for both one and three minutes. Two percent (2 %) malt extract agar (MEA) and cellulose Czapek agar (CCA) was used to recover general fungi and cellulose-loving fungi, respectively. Non-culturable air samples were collected at the aforementioned sampling locations. Indoor samples were collected for ten minutes and outdoor samples were collected for both five and ten minutes. Outdoor air samples were collected near the entrance of the building. Temperature and relative humidity measurements were collected from each air sampling location by a battery operated, direct readout Hygroskopâ meter.

Contact Plate Samples

To determine fungal burden on horizontal and vertical surfaces of these rooms, five to eight contact plate samples were collected from each room. Samples were collected from randomly selected horizontal and vertical surfaces. Sampling was conducted by pressing the MEA-filled Rodacâ plate against the surface of interest for five seconds. A total of 13 contact plate samples were collected.

Swab Samples

Swab samples were collected from surfaces of each supply diffusers and return troughers in each room. They were collected by wiping a known area of surface with a sterile cotton swab (Culturette®) wetted with holding media. Approximately 5 in^2 area was wiped for return trougher and 4 in^2 for supply diffusers. One sample was collected from an exhaust grill in room 10817. The swab was then placed directly into its holder. Each holder was labeled with an identifiable number. A total of 12 wipe samples were collected from these rooms.

Vacuum Dust Samples

Dust accumulated on carpeting, chairs and fabric system furniture, and the plenum were collected with a High Efficiency Particulate Air (HEPA) vacuum attached with a special "sock" device. For each carpet sample, a 3-ft by 3-ft area was vacuumed for at least five minutes. Total surface areas of 9 ft^2 were vacuumed from system furniture and chairs, and composite as one sample. Dust accumulated above the ceiling plenum was also vacuumed and composite as one sample. One composite furniture sample and one composite plenum sample were collected from each room. One carpet sample was collected from room 10817.

All samples collected were sent for next morning delivery to FOH's Environmental Microbiology Laboratory (EML) in Philadelphia, Pennsylvania for analysis.

Laboratory Procedures

Upon receipt, all Andersen® air and contact plate samples were incubated in a 25˚C incubator. Each swab sample was suspended in sterile distilled water, diluted serially, and inoculated onto agar plates. Both MEA and CCA were used for retrieving fungi. At least three dilution series were used for each sample. Each vacuum dust sample was sieved through a 250 mm sieve. The fine dust (< 250 mm) retrieved was then weighed and followed the dilution plating for fungal analysis.

All plates were incubated in a 25˚C incubator. They were examined every other day for up to 10 days to ensure the full recovery of fungi. Fungal identification was based on colony morphology, spores and conidia formation. Total fungal colonies formed on each MEA plate and *Stachybotrys chartarum* on CCA plates were counted and recorded. Fungal levels in samples were presented as colony forming units (CFUs) per measuring unit. For example, CFU/m^3 for Andersen® air samples, CFU/in^2 for wipe samples, CFU/plate for contact plate samples, and CFU/g of fine dust for vacuum dust samples.

All Zefonâ cassette samples were analyzed by the Environmental Microbiology Laboratory in Escondido, California for direct microscopic examination. Fungal spores were identified and their airborne levels were presented as spores/m^3.

RESULTS AND DISCUSSION

Temperature and Relative Humidity

Indoor temperature and relative humidity measurements ranged from 69.9°F to 73.5°F, and 16.7% – 19.4%, respectively (Table 1). Outdoors temperature reading was lower (47.5°F), but with a slightly higher relative humidity (20.9%) (Table 1).

Microbiological Analyses Results

All laboratory analytical results from FOH's EML are presented in Attachment A in a laboratory report #NOAA-00-32R. Results from microscopic examination of Zefonâ cassette samples are presented in Attachment B.

Air Samples

Andersen Results

Outdoor airborne fungal levels were higher than those of indoors (Table 1). *Cladosporium* dominated outdoor fungal flora. Other fungi recovered from outdoors were *Penicillium*, *Epicoccum*, *Paecilomyces*, and Basidiomycetes. Fungi detected indoors were *Alternaria* and *Cladosporium*. *Stachybotrys chartarum* was not detected from these samples.

Zefon Results

Outdoor fungal spore levels were higher than those of indoors (Table 1). Fungal spore types recovered from outdoors were *Cladosporium* and Basidiomycetes. Fungal spore types recovered from indoors were *Cladosporium*, *Penicillium*/*Aspergillus* types, and Smuts, Periconia, Myxomycetes. *Stachybotrys chartarum* was not detected from any sample collected.

Table 1. Temperature and relative humidity measurements and airborne fungal levels at different rooms of the 10th floor in SSMC-3 on February 17, 2000.

Rooms / Parameters	10817 Reference[#]	10129	Outdoors
Temperature (°F)	69.9	73.5	47.5
Relative Humidity (%)	19.4	16.7	20.9
Airborne Fungal Levels (CFU/m^3)	12	12	200* 212
Total Fungal Spores ($Spores/m^3$)	26	27	294* 280

[#] Indoor reference.

* Two samples were collected from outdoors.

Swab Samples

Most (8 out of 11) samples collected from surfaces of supply diffusers and return troughers in light fixtures were below the detection limits (BDL) (3 CFU/in^2 for supply diffuser and 2 CFU/in^2 for return trougher). Samples showing fungal growth had low fungal levels (2 CFU/in^2 to 8 CFU/in^2). The sample collected from an exhaust grill in 10817 showed 13 CFU/in^2 of fungal growth with *Cladosporium* as the predominant fungi. *Stachybotrys chartarum* was not detected from these samples.

Contact Plate Samples

In general, higher fungal levels were detected from the horizontal surfaces than vertical surfaces (Table 2). Fungal levels on vertical surfaces ranged from BDL of 1 CFU/plate to 2 CFU/plate. Fungal levels on horizontal surfaces ranged from BDL of 1 CFU/plate to 30 CFU/plate. Higher fungal levels were detected in room 10129 than those of room 10817. Fungi recovered from these samples were *Cladosporium*, *Penicillium*, *Aspergillus niger*, *Alternaria*, *Epicoccum*, *Rhizopus*, *Mucor*, and Basidiomycetes. *Stachybotrys chartarum* was not detected from these samples.

Table 2. Fungal levels (CFU/plate) on horizontal and vertical surfaces of different rooms at the 10th

floor of SSMC-3, by contact plate sampling collected on February 17, 2000.

Rooms	**10817** Reference[#]	**10129**
Horizontal Surfaces (CFU/plate)	1 (1**)	4 – 30* (4)
Vertical Surfaces (CFU/plate)	<1 (4)	<1 – 2 (4)

[#] Indoor reference. * Ranges. ** Total sample number.

Vacuum Dust Samples

Stachybotrys chartarum was not detected from these samples (Table 3).

Table 3. Total fungal levels (CFU/g of fine dust) in fine dust collected from carpet, plenum, and furniture of rooms 10129 and 10817 of SSMC-3, by vacuum dust sampling, collected on February 17, 2000.

Rooms	**10817** **Reference[#]**	**10129**
Plenum (CFU/g of fine dust)	122,772 (-*)	12,277 (-)
Carpet (CFU/g of fine dust)	9,505 (-)	NA**
Furniture (CFU/g of fine dust)	7,525 (-)	2,000 (-)

[#] Indoor reference.

* +: *Stachybotrys chartarum* was detected on MEA and/or CCA plates.

-: *Stachybotrys chartarum* was not detected on MEA and CCA plates.

** Not applicable.

Carpet Dust

The fungal level in the fine dust collected from carpeting of room 10817 was at 9,505 CFU/g of fine dust level (Table 3). *Aureobasidium* dominated this sample, followed by *Penicillium, Paecilomyces,* and *Cladosporium.*

Furniture Dust

Fungal levels in the fine dust in furniture of these rooms were at the levels of 10^3 CFU/g of fine dust (Table 3). Predominant fungi detected were *Penicillium* dominated the sample collected from the reference room, 10817. Other fungi recovered, in a descending order, were *Cladosporium, Aureobasidium, Alternaria, Aspergillus niger, Paecilomyces,* and *Trichoderma*. *Aureobasidium, Penicillium, Paecilomyces,* and yeast were recovered from furniture dust sample collected from room 10129.

Plenum Dust

The fungal level in the fine dust in plenum of indoor reference room, 10817, was higher than that of room 10129 (10^5 vs. 10^4 CFU/g of fine dust) (Table 3). *Penicillium* and *Aspergillus* (*Aspergillus niger* included) dominated these samples. Other fungi recovered were, *Cladosporium, Alternaria, Paecilomyces,* and *Trichoderma*.

CONCLUSIONS

· Indoor airborne fungal levels, by Andersen sampling, and indoor spore levels, by Zefon sampling, were lower than those of outdoors.

· *Stachybotrys chartarum* was not detected from any air, swab, contact plate, and dust samples.

· In general, fungal burden on vertical surfaces was lower than that of horizontal surfaces.

· Higher fungal levels were detected from samples collected from horizontal surfaces in room 10129 than those in reference room 10817.

· Very low fungal burden was detected from wipe samples collected from surfaces of supply diffusers and return troughers in light fixture.

· The fungal level in plenum dust of reference room was higher than that of room 10129 (10^5 vs. 10^4 CFU/g of fine dust). *Penicillium* and *Aspergillus* (*Aspergillus niger* included) dominated these samples.

· Fungal levels in furniture dust of these rooms were at 10^3 CFU/g levels.

RECOMMENDATIONS

· Conduct a thorough cleaning of these rooms by HEPA vacuuming and wet wiping.

· Conduct any above ceiling plenum work after hour. Thoroughly HEPA vacuum the surrounding areas afterwards.

· Implement an emergency water intrusion protocol for this building to adequately manage any unexpected water intrusion in order to prevent fungal proliferation.

ATTACHMENT A

Microbiological laboratory report #NOAA-00-32R for samples collected

from tenth floor of SSMC-3, on February 17, 2000.

ATTACHMENT B

Results from microscopic examination of Zefon air samples collected

from tenth floor of SSMC-3, on February 17, 2000.

USPHS DFOH ENVIRONMENTAL MICROBIOLOGY LABORATORY, PHILADELPHIA, PA

LABORATORY REPORT #NOAA-00-32R

Client agency: National Oceanic and Atmospheric Administration, Silver Spring, MD

POIS#/task #: D8H00CO31200 / 9903

Sampling date: 2/17/00

Dates of inoculation: 2/17/00 (airs and contact plates), 2/18/00 (wipes), and 2/20/00 (dust)

General location: SSMC-3, Silver Spring, MD

Specific location: 10th floor

Sampling techniques: Air (Andersen N-6 sampler), contact plate, wipe, and vacuum dust samplings

Medium used: Malt extract agar (MEA) and cellulose Czapek agar (CCA) for fungi

Samples submitted by: J. Sobelman

Date characterization completed: 3/1/00

(A) Air samples on MEA and CCA plates

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
3-10817-0217A1, A2	10th floor, room 10817, small conference room	84.9	1. *Alternaria* (1*) CFU/m^3 = 12	No
3-10129-0217A1, A2	10th floor, room 10129, center of office	84.9	1. *Cladosporium* (1) CFU/m^3 = 12	No
3-OA1-0217, 3-OA2-0217	Outside bldg. 3	84.9	1. *Cladosporium* (13) 2. *Penicillium* (2) 3. *Epicoccum* (1) 4. *Paecilomyces* (1) CFU/m^3 = 200	No

3-OA3-0217, 3-OA4-0217	Outside bldg. 3	28.3	1. *Cladosporium* (5) 2. Basidiomycetes (1) CFU/m^3 = 212	No
FB	Field blank	NA#	No fungal growth	No

Sample ID	Sampling Location	Air Volume (L)	Fungi on MEA @ 25º C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
SB	Shipping blank	NA	No fungal growth	No

(B) Contact plate samples on MEA plates

Sample ID	Sampling Location	Fungi detected on MEA @ 25º C
3-10817-0217CP1	10th floor, room 10817, S wall near door	**No fungal growth** CFU/plate < 1
3-10817-0217CP2	10th floor, room 10817, N wall near window	**No fungal growth** CFU/plate < 1
3-10817-0217CP3	10th floor, room 10817, E wall	**No fungal growth** CFU/plate < 1
3-10817-0217CP4	10th floor, room 10817, W wall	**No fungal growth** CFU/plate < 1
3-10817-0217CP5	10th floor, room 10817, table	1. *Cladosporium* (1) **CFU/plate = 1**
3-10129-0217CP1	10th floor, room 10129, S wall	**No fungal growth** CFU/plate < 1

3-10129-0217CP2	10th floor, room 10129, N wall	1. *Cladosporium* (1) 2. *Penicillium* (1) **CFU/plate = 2**
3-10129-0217CP3	10th floor, room 10129, E wall	1. *Aureobasidium* (1) **CFU/plate = 1**
3-10129-0217CP4	10th floor, room 10129, W wall	**No fungal growth** CFU/plate < 1

Sample ID	**Sampling Location**	**Fungi detected on MEA @ 25º C**
3-10129-0217CP5	10th floor, room 10129, top of desk	1. *Cladosporium* (2) 2. *Penicillium* (1) 3. Basidiomycetes (1) **CFU/plate = 4**
3-10129-0217CP6	10th floor, room 10129, top of system furniture	1. *Cladosporium* (4) 2. *Alternaria* (3) 3. *Epicoccum* (1) 4. *Penicillium* (1) **CFU/plate = 9**
3-10129-0217CP7	10th floor, room 10129, top of big cabinet	1. *Penicillium* (14) 2. *Cladosporium* (10) 3. *Aspergillus niger*** (3) 4. *Alternaria* (2) 5. *Rhizopus* (1) **CFU/plate = 30**

3-10129-0217CP8	10th floor, room 10129, floor	1. *Cladosporium* (2) 2. *Aspergillus niger*** (1) 3. *Mucor* (1) 4. *Penicillium* (1) **CFU/plate = 5**

(C) Wipe samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
Blank	Blank	Blank	NA	10X-MEA 10X-CCA	No fungal growth	No

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
W25	3-10817-0217R1	10th floor, room 10817, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W26	3-10817-0217R2	10th floor, room 10817, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W27	3-10817-0217R3	10th floor, room 10817, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W28	3-10817-0217R4	10th floor, room 10817, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No
W29	3-10817-0217S1	10th floor, room 10817, supply	4	10X-MEA 10X-CCA	1. *Rhizopus* (1) CFU/in^2 = 3	No
W30	3-10817-0217S2	10th floor, room 10817, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

W31	3-10817-0217S3	10th floor, room 10817, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No
W32	3-10817-0217S4	10th floor, room 10817, supply	4	10X-MEA 10X-CCA	1. *Alternaria* (1) 2. *Cladosporium* (1) 3. *Penicillium* (1) CFU/in^2 = 8	No
W33	3-10817-0217E1	10th floor, room 10817, exhaust	4	10X-MEA 10X-CCA	1. *Cladosporium* (3) 2. *Penicillium* (1) 3. yeast (1) CFU/in^2 = 13	No
W34	3-10129-0217R1	10th floor, room 10129, return	5	10X-MEA 10X-CCA	1. *Cladosporium* (1) CFU/in^2 = 2	No
W35	3-10129-0217R2	10th floor, room 10129, return	5	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 2	No

FOH ID	Sample ID	Sampling Location	Area (in^2)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C
W36	3-10129-0217S1	10th floor, room 10129, supply	4	10X-MEA 10X-CCA	No fungal growth CFU/in^2 < 3	No

(D) Vacuum dust samples on MEA and CCA plates

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25º C

V05	3-10817-0217V01	10th floor, room 10817, furniture	0.101##	40X-MEA 10X-CCA	**1. *Penicillium* (17)** 2. *Cladosporium* (13) 3. *Aureobasidium* (3) 4. *Alternaria* (2) 5. *Aspergillus niger*** (1) 6. *Paecilomyces* (1) 7. *Trichoderma* (1) **CFU/g = 7,525**	No
V06	3-10817-0217V02	10th floor, room 10817, carpet	0.101	40X-MEA 10X-CCA	**1. *Aureobasidium* (13)** 2. *Paecilomyces* (2) 3. *Penicillium* (2) 4. *Cladosporium* (1) 5. yeast (6) **CFU/g = 9,505**	No

FOH ID	Sample ID	Sampling Location	Weight (g)	Dilution factor	Fungi on MEA @ 25°C	Presence of *Stachybotrys chartarum**** on CCA @ 25° C

V07	3-10817-0217V03	10th floor, room 10817, above ceiling	0.101	400X-MEA 10X-CCA	**1. *Penicillium* (26)** 2. *Alternaria* (2) 3. *Aspergillus niger*** (1) 4. *Aspergillus sp.* (1) 5. *Cladosporium* (1) CFU/g = 1.2×10^5	No
V08	3-10129-0217V01	10th floor, room 10129, furniture	0.029§	40X-MEA 10X-CCA	**1. *Aureobasidium* (1)** 2. *Penicillium* (1) 3. *Pithomyces* (1) 4. yeast (2) **CFU/g = 2,000**	No
V09	3-10129-0217V02	10th floor, room 10129, above ceiling	0.101	40X-MEA 10X-CCA	**1. *Aspergillus niger*** (9)** 2. *Cladosporium* (8) 3. *Penicillium* (8) 4. *Paecilomyces* (5) 5. *Trichoderma* (1) CFU/g = 1.2×10^4	No

* Colony counts. ** Opportunistic fungi. *** Toxigenic fungi.

Not applicable.

5ml of sterilized distilled water were added instead of 10ml.

§ Equivalent amounts of sterilized distilled water were added instead of 10ml.